

REMARKS

Applicant intends this response to be a complete response to the Examiner's 25 May 2010 Non-Final Office Action. Applicant has labeled the paragraphs in his response to correspond to the paragraph labeling in the Office Action for the convenience of the Examiner.

DETAILED ACTION

The Examiner states and contends as follows:

1. Response filed 25 February 2010 to the Office Action mailed 02 February 2010 is acknowledged and entered.

Applicants acknowledge this statement.

Claims Status

The Examiner states and contends as follows:

2. Claims 1-30 are currently pending.

Applicants acknowledge this statement.

Restriction/Election

The Examiner states and contends as follows:

3. Applicants' response and election with traverse of Group II encompassing Claims 14-30 drawn to a multistep method for further prosecution filed 25 February 2010 to the election requirement in Office Action mailed 02 February 2010 is acknowledged and entered. The traversal is on the grounds that the inventions grouped as I-II should be rejoined (See, Response filed 25 February 2010, Page 10, Lines 2-7).

- (i) Applicants are of the opinion that the 2 Groups should be rejoined and examined as one invention, because:
- (ii) the method utilizes the composition and is non-functional in absence of composition (See, Response filed 25 February 2010, Page 10, Lines 2-3);
- (iii) the two groups share the most common feature (See, Response filed 25 February 2010, Page 10, Lines 3-4); and
- (iv) "MPEP guidelines argues against restriction for the interconnectedness" as is present in Claims encompassing Groups I-II inventions (See, Response filed 25 February 2010, Page 10, Lines 6-7).

The two inventions do not share a common technical feature, where said technical feature of inter-relatedness between the two inventions is a CONTRIBUTION OVER THE PRIOR ART (37 C.F.R. §1.475).

As is of record in the Office Action mailed 02 February 2010 (Pages 2-6), group I composition is a culture medium comprising certain concentration of cations and percentage of saline (i.e., NaCl) to support bacterial growth which is well known in the relevant art (see for e.g., U.S. Patent 6,077,429, Abstract, Figures 1-3, Column 9, Line 55-Column 10, Line 37). The special technical feature of Group II is a method to load a bioreactor with a pollutant containing stream comprising brine or wastewater and bacteria that degrade the pollutants, which again is well known in the relevant art (see, e.g., U.S. Patent 6,077,429 as indicated above and additionally, Column 10, Line 38 to Column 11, References 1, 5, 7-15 and 23) and Okeke et al., (2002. Reduction of perchlorate and nitrate by salt tolerant bacteria. Environmental Pollution, Volume 118, Pages 357-363), Abstract. Page 358, Column 2, Lines 4-37; Page 358, Column 2, Line 4 to Page 359, Column 2, Line 49; Page 360, Column 1, Lines 2-37; Page 360, Column 2, Lines 1-2 and 16-36 and Figures 1-2). Accordingly, the two inventions do not share a common technical feature according to 37 C.F.R.

§ 1.475; MPEP §1850 [R-7]. Burden is not an issue when lack of unity is established.

Applicants' arguments regarding the restriction requirements in the response filed 25 February 2010 to Office Action mailed 02 February 2010 have been carefully and fully considered but are not found persuasive because of the reasons of record at pages 2-5, items 6-9 of the Office Action mailed 02 February 2010; and for additional reasons discussed *supra*. Therefore the restriction requirement in the Office Action mailed 02 February 2010 is still deemed proper and is made FINAL.

Accordingly, Claims 1-13 are withdrawn from further consideration as being directed to a non-elected invention. See 37 C.F.R. §1.142(b) and M.P.E.P. §821.03.

While Applicants still strongly assert that the restriction requirement is improper due to the close nexus between the two groups, Applicants acknowledge the Examiner's decision to make the restriction final.

The Examiner states and contends as follows:

4. Claims 14-30 are examined on merits.

Applicants acknowledge this statement.

Priority

The Examiner states and contends as follows:

5. Claim for domestic priority under 35 U.S.C. §119(e) to Provisional U.S. Application Serial Number 60/523,637 filed 20 November 2003 is acknowledged.
6. Claim for priority under 35 U.S.C. §371 to PCT/US04/38808 filed 19 November 2004 is acknowledged.

Applicants acknowledge the Examiner's statements.

References Cited in 371 Application

The Examiner states and contends as follows:

7. The references cited in the Search Report filed 18 May 2006 as "Documents submitted with 371 Application" have been considered, but will not be listed on any patent resulting from this application because they were not provided on a separate list in compliance with 37 C.F.R. §1.98(a)(1). In order to have the references printed on such resulting patent, a separate listing, preferably on a PTO/SB08A and 08B form, must be filed within ONE MONTH of the mailing date of this communication. NO EXTENSION OF TIME WILL BE GRANTED UNDER EITHER 37 C.F.R. §1.136(a) OR (b) to comply with this requirement.

Applicants acknowledge the Examiner's statements and have submitted an IDS previous to this response, within the stated 1 month timer. Applicants believe that their recent IDS filing renders this aspect of the Office Action Moot.

Specification Objected

The Examiner states and contends as follows:

8. At page 1, first paragraph 1, the Application priority data regarding Applicants' Claim for priority to PCT/US04/38808 filed 19 November 2004 is lacking. Appropriate correction is required.

Applicants are not sure about this objection. Applicants submitted a preliminary amendments on 18 May 2006, correcting the priority claims. That version is set forth above.

Claim Rejections 35 U.S.C. §112
35 U.S.C. §112, second paragraph of

10. **Claims 30** stands rejected under 35 U.S.C. § 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

The Examiner states and contends as follows:

The limitation, "first and second ion exchange resins" in Claim 30 lacks insufficient antecedent basis for said limitation in the cited claim, because Claim 27 from which Claim 30 depends does not recite two different ion exchange resins. Appropriate correction is required.

Applicants have amended claim 30 to correct its dependency. Claim 30 should have depended on claim 29 instead of claim 27. Applicants, therefore, respectfully request withdrawal of this rejection, which can have no estoppel effect as it merely corrects an error in claim dependency.

Claim Rejections - 35 U.S.C. §102/103

14. **Claim 14-26** stand rejected under 35 U.S.C. §102(b) as anticipated by or, in the alternative, under 35 U.S.C. § 103(a) as obvious over Logan (U.S. Patent 6,214,607 B1).

The Examiner states and contends as follows:

Claims 14-26 are drawn to a method to degrade at least one contaminant present in a contaminated brine stream when said stream is:

- ▲ fed to a bioreactor containing a mixed bacterial culture, wherein;
- ▲ said bacterial culture degrades under anoxic/anaerobic conditions said at least one pollutant in said brine stream;
- ▲ to said bioreactor is also added an effective quantity of a divalent cation precursor in such amount that said divalent cation quantity maintains a divalent to monovalent cation molar ratio at ≥ 0.05 ;
- ▲ said degradation of said brine stream contaminant takes place at a temperature for a duration to bring the contaminant concentration at a desired level, while divalent to monovalent cation molar ratio is maintained at ≥ 0.05 ;
- ▲ said bioreactor is sealed to eliminate/reduce oxygen;
- ▲ said reactor being sparged with an oxygen-free gas selected from nitrogen (N₂) or Argon (Ar) or mixtures thereof;
- ▲ divalent cation selected from soluble salt of:
 - Ba²⁺, Ca²⁺, Mg²⁺, Sr²⁺; or
 - Ca²⁺, Mg²⁺, Sr²⁺; or
 - Ca²⁺ and Mg²⁺; or
 - is Mg²⁺;
- ▲ the contaminant is selected from the group perchlorate (ClO₄⁻) or nitrate (NO₃⁻) or mixture thereof;
- ▲ nutrients comprising an inorganic, or organic nutrient source in an amount greater than a stoichiometric amount of electron required to reduce the ClO₄⁻ and/or NO₃⁻ contaminant present in the brine solution for sustained microbial growth;
- ▲ the inorganic energy source is selected from the group H₂ gas, a H₂ delivery chemical or mixtures thereof;
- ▲ the organic nutrients are selected from the group consisting of acetate, ethanol, lactate,

methanol, or mixtures thereof; and

- the contaminated brine solution is a ClO_4^- and/or NO_3^- contaminated ion-exchange regenerate brine.

Regarding Claims 14-26, Logan teaches degradation of ClO_4^- in a brine solution, comprising per liter of deionized water: 5.85 mg of NaCl, 6.5 mg of MgSO_4 , 4.3 mg of $\text{CaSO}_4 \cdot 2\text{H}_2\text{O}$ and 48.0 mg of CaCO_3 with 100 mg acetate, nitrogen and phosphorus (Example 1; Column 8, Lines 40-45). Logan further teaches that degradation studies for said ClO_4^- contained in said brine solution was carried out in crimp-topped serum vials (*i.e.*, bioreactors), whose head space was purged with N_2 gas, said bioreactors inoculated with ClO_4^- degrading mixed microbial consortium and the degradation was carried out for 7 days at room temperature. In 4 days, 39% ClO_4^- was removed (Example 1; Column 8, Lines 25-40). Subsequently column reactors were inoculated with the same microbial consortium wherein the column reactors contained the same ClO_4^- -containing brine solution described *supra* (Example 1; Column 8, Lines 40-45). Logan further teaches that the pH of the column was in range of 5.0-8.0, the column temperature 10°C to 30°C to keep bacterial cells viable and active, and the system is comprised of an "oxidizable substrate serving as electron donor. Oxidizable substrates include acetate, ethanol, methanol and hydrogen gas (Column 6, Lines 8-20). Based on the concentrations of each of NaCl, MgSO_4 , $\text{CaSO}_4 \cdot 2\text{H}_2\text{O}$ and CaCO_3 said brine solution has a divalent to monovalent cation ratio of 0.1060. Thus, Lawrence et al., teach each and every limitation: H_2 gas as an inorganic nutrient, acetate, methanol, or ethanol as an organic nutrient, purging with N_2 gas, ClO_4^- -contaminated brine solution having divalent to monovalent cation ratio of 0.1060 (*i.e.*, ≥ 0.05), and 39% degradation of ClO_4^- of the invention claimed in Claims 14-26.

Therefore, the reference is deemed to anticipate the cited claims.

However, even if the reference and the claimed method are not one and the same and there is, in fact, no anticipation, the reference method would, nevertheless, have rendered the claimed method obvious to one of ordinary skill in the art at the time the claimed invention was made in view of the fact that the reference teaches same materials and same steps. The only difference between the reference method and the instantly claimed invention resides in the concentrations of contaminant. Nevertheless, the reference method (*i.e.*, Logan) teaches each and every component and each of the method steps as instantly claimed and within the range of the materials instantly claimed. Additionally, if there is any guidance in the reference regarding the concentration range of a material as is presented for each one of cations (*e.g.*, Ca^{2+} and Mg^{2+}) and ClO_4^- from Logan; the adjustment of particular conventional working equivalents (*e.g.*, concentration) is deemed merely a matter of judicious selection and routine optimization of a result-effective parameter that is well within the purview of the skilled artisan for which the Examiner -cited reference has provided some guidance.

Thus, the claimed invention as a whole was clearly *prima facie* obvious especially in the absence of sufficient, clear and convincing evidence to the contrary.

From the teachings of the reference cited *supra*, it is apparent that one of ordinary skill in the art would have had a reasonable expectation of success in producing the claimed invention. Therefore, the invention as a whole was *prima facie* obvious to one of ordinary skill in the art at the time the invention was made, as evidenced by the references, especially in the absence of evidence to the contrary.

Applicants first note that Logan makes no mention of brines. In fact, the word brine never even appears in Logan. The Examiner seems to equate water that has any amount of dissolved ions to a brine. This conclusion is simply incorrect. Seawater is a quintessential brine. Sea water is a 3.5% NaCl brine, with other metal ions present at much lower concentrations. On the other hand, free water has a salinity of 100 ppm NaCl. In fact, the following table available from Wikipedia (<http://en.wikipedia.org/wiki/Salinity>) clearly shows that brines have a NaCl content of that of sea water or above:

Water Salinity			
Fresh water	Brackish water	Saline water	Brine
< 0.05 %	0.05 – 3 %	3 – 5 %	> 5 %
< 0.5 ‰	0.5 – 30 ‰	30 – 50 ‰	> 50 ‰

While Logan does utilize a divalent to monovalent ratio of ≥ 0.05 , Logan is directed at taking chlorate or perchlorate out of **fresh water** sources to make **drinking water**. Logan does not disclose, teach or even lead one of ordinary skill in the art to look at microbial stability in brines having a salinity of at least 3%.

Moreover, Applicants believe that the Examiner miss calculated the M^{2+}/M^{1+} ratio for Logan. Logan teaches an aqueous solution having: 5.85 mg of NaCl, 6.5 mg of $MgSO_4$, 4.3 mg of $CaSO_4 \cdot 2H_2O$ and 48.0 mg of $CaCO_3$, which give rise to a M^{2+}/M^{1+} of 9.26 as calculated in the spreadsheet below.

Chemical	Concentration (mg/L)	mol/mg	mol/L	Mass cation	divalent sum	ratio
NaCl	5.85	1.724E-05	0.0001009	0.00232		
MgSO4	6.5	8.333E-06	5.417E-05	0.0013	0.0215	9.27
CaSO ₄ •2 H ₂ O	4.3	5.814E-06	0.000025	0.001		
CaCo3	48	0.00001	0.00048	0.0192		

Clearly, 9.26 is greater than 0.05%; in fact, it is orders of magnitude greater than 0.05%. While this still meets the claims limitation, the real truth of the matter, the solution of Logan are simply not brines having only milligrams of salts dissolved in water. In brines, the major ions are present in gram quantities.

The present invention is related directly to using microbes to degrade contaminants in brines, solutions having a salinity greater than or equal to 3%. The present invention relates to how to stabilize microorganisms from salt toxicity at high salinity – salinity of 3% or higher. Logan does not even mention salt toxicity so no one would not look to Logan for a disclosure regarding brines as fresh water is the antithesis of a brine.

Because Logan does not even mention brines and because Logan does not disclose, teach or even lead one of ordinary skill in the art to look at microbial stability in brines, Logan cannot not anticipate the claims of this invention. Applicants, therefore, respectfully request withdrawal of this rejection, which can have no estoppel effect as it merely defines a term clearly defined in the application.

Moreover, Logan cannot even render these claims obvious as Logan does not even mention brines nor does anything in Logan look to stabilizing microbes in salt water as Logan is directed at making drinking water not stabilizing brines – waters that are not drinking waters under in definition. In fact, in a further work, (Logan, Wu and Unz, 2001, Biological Perchlorate Reduction in High Salinity Solutions, Water Research) Logan does study a brine and makes no reference to adding magnesium (or any divalent ion) to attain a ratio of divalent to monovalent ratio. In this work Logan increases the sodium concentration in water and demonstrates a decrease in the ability of his culture to degrade perchlorate. The work does not report any attempts to increase salt tolerance by altering any of the other ion concentrations.

Claim Rejections - 35 U.S.C. § 103

16. **Claims 27-30** stand rejected under 35 U.S.C. §103 (a) as obvious over the combined teachings from Logan (U.S. Patent 6,214,60781) in view of Venkatesh et al (U.S. Patent 6,066,257).

The Examiner states and contends as follows:

Claims 27-30 are drawn to a method to degrade at least one contaminant present in a contaminated brine stream when said stream is:

fed to a bioreactor containing a mixed bacterial culture, wherein;
said bacterial culture degrades under anoxic/anaerobic conditions said at least one pollutant in said brine stream;
the column resin in said reactor is exhausted, or is not able to remove the contaminant, the flow of contaminant-containing brine is stopped to said reactor,
or fed to a second reactor, while the resin in the first column is recharged with the addition of a divalent cation solution to maintain the divalent to monovalent cation molar ratio at ≥ 0.05 .

Regarding Claims 27-30, teachings from Logan have been discussed *supra*. Logan however, is silent regarding switching the reactors or regeneration of column resin.

Venkatesh et al., teach that the units employed in Venkatesh et al's method steps have a regeneration zone, wherein the low concentration brine is used as regenerant. Then the waste stream comprising perchlorate containing brine is treated in a bioreactor and the resulting stream is recycled (Abstract). Thus, Venkatesh et al' method remedies the discrepancy of column/resin regeneration step in Logan's method.

One having ordinary skill in the art at the time of the claimed invention would have been motivated to modify/combine the teachings from Logan with those of Venkatesh et al., to obtain a method wherein the column/resin incapacitated of removing the perchlorate contaminant from the brine would be regenerated; because Venkatesh et al., teach regeneration of columns/resins and refeeding the perchlorate-contaminated brine to said regenerated columns/resins.

It would have been *prima facie* obvious to a person of ordinary skill in the art at the time the claimed invention was made to combine the teachings from Logan with those of Venkatesh et al., to obtain a method wherein the column/resin incapacitated of removing the perchlorate contaminant from the brine would be regenerated; because Venkatesh et al., teach regeneration of columns/resins and refeeding the perchlorate-contaminated brine to said regenerated columns/resins. The recited prior art may be silent regarding the exact technique as are instantly claimed, however, since similar steps and same components and principle are described in the prior art references, application of those parameters and steps is deemed merely a matter of judicious design selection of a result-effective

